

Phylogenetic tree of human fibroblast growth factor (hFGF) genes. The tree shows the evolutionary relationships between various hFGF isoforms. The top cluster includes FGF23, hFGF-19, hFGF-21, hFGF-17, hFGF-8, and hFGF-18. The middle cluster includes hFGF-10, hFGF-7, hFGF-22splice, hFGF-3, hFGF-16, hFGF-9, hFGF20, hFGF-1, and hFGF-2. The bottom cluster includes hFGF-11, hFGF-12, hFGF-14, hFGF-15, hFGF-13, hFGF-4, hFGF-6, and hFGF-5 pLTR122. The tree is rooted on the left and branches out to the right.

- FGF23
- hFGF-19
- hFGF-21
- hFGF-17
- hFGF-8
- hFGF-18
- hFGF-10
- hFGF-7
- hFGF-22splice
- hFGF-3
- hFGF-16
- hFGF-9
- hFGF20
- hFGF-1
- hFGF-2
- hFGF-11
- hFGF-12
- hFGF-14
- hFGF-15
- hFGF-13
- hFGF-4
- hFGF-6
- hFGF-5 pLTR122

FIGURE 1

Mouse FGF-23

10 20 30 40 50 60
 ATGCTAGGGACCTGCCCTAGACTCCTGGTGGGCGTGCTCTGCACTGTCTGCAGCTTGGGC
 M L G T C L R L L V G V L C T V C S L G
 70 80 90 100 110 120
 ACTGCTAGAGCCTATCCAGACACTTCCCCATTGCTTGGCTCCAACTGGGGAAGCCTGACC
 T A R A Y P D T S P L L G S N W G S L T
 130 140 150 160 170 180
 CACCTGTACACGGCTACAGCCAGGACCAGCTATCACCTACAGATCCATAGGGATGGTCAT
 H L Y T A T A R T S Y H L Q I H R D G H
 190 200 210 220 230 240
 GTAGATGGCACCCTCATCAGACCATCTACAGTGCCTGATGATTACATCAGAGGACGCC
 V D G T P H Q T I Y S A L M I T S E D A
 250 260 270 280 290 300
 GGCTCTGTGGTGATAACAGGAGCCATGACTCGAAGGTTCCCTTGTATGGATCTCCACGGC
 G S V V I T G A M T R R F L C M D L H G
 310 320 330 340 350 360
 AACATTTTGGATCGCTTCACTTCAGCCCAGAGAATTGCAAGTTCGCCAGTGGACGCTG
 N I F G S L H F S P E N C K F R Q W T L
 370 380 390 400 410 420
 GAGAATGGCTATGACGTCTACTTGTCCGAGAAGCATCACTACCTGGTGAGCCTGGGCCGC
 E N G Y D V Y L S Q K H H Y L V S L G R
 430 440 450 460 470 480
 GCCAAGCGCATTTTCCAGCCGGGCACCAACCCGCCGCCCTTCTCCCAGTTCCTGGCTCGC
 A K R I F Q P G T N P P P F S Q F L A R
 490 500 510 520 530 540
 AGGAACGAGGTCCCCTGCTGCACTTCTACACTGTTCCGCCACGGCGCCACACGCGCAGC
 R N E V P L L H F Y T V R P R R H T R S
 550 560 570 580 590 600
 GCCGAGGACCCACCCGAGCGGACCCACTGAACGTGCTCAAGCCGCGGCCCCCGCGCCACG
 A E D P P E R D P L N V L K P R P R A T
 610 620 630 640 650 660
 CCTGTGCTGTATCCTGCTCTCGCGAGCTGCCGAGCGCAGAGGAAGGTGGCCCCCGCAGCC
 P V P V S C S R E L P S A E E G G P A A
 670 680 690 700 710 720
 AGCGATCCTCTGGGGGTGCTGCGCAGAGGCCGTGGAGATGCTCGCGGGGGCGCGGGAGGC
 S D P L G V L R R G R G D A R G G A G G
 730 740 750 760
 GCGGATAGGTGTGCGCCCTTTCCAGGTTCTGTCTAG
 A D R C R P F P R F V

FIGURE 2

00001598-03094

FIGURE 3

Human FGF-23

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      10      20      30      40      50      60
atgttgggggcccgcctcagggtcttgggtctgtgccttgtgcagcgtctgcagcatgagc
H L G A R L R L W V C A L C S V C S M S

      70      80      90     100     110     120
gtcctcagagcctatcccaatgcctccccactgctcggctccagctggggtggcctgac
V L R A Y P N A S P L L G S S W G G L I

      130     140     150     160     170     180
cacctgtacacagccacagccaggaacagctaccacctgcagatccacaagaatggccat
H L Y T A T A R N S Y H L Q I H K N G H

      190     200     210     220     230     240
gtggatggcgcccccctcagaccatctacagtgccttgatgatcagatcagaggatgct
V D G A P H Q T I Y S A L M I R S E D A

      250     260     270     280     290     300
ggctttgtggtgattacaggtgtgatgagcagagatocctctgcattggatttcagaggc
G F V V I T G V M S R R Y L C M D F R G

      310     320     330     340     350     360
aacatttttggtacacactatttcgacccggagaaactgcagggttccaacaccagacgtg
N I F G S H Y F D P E N C R F Q H Q T L

      370     380     390     400     410     420
gaaaocgggtacgacgtctaccactctcctcagtatcacttcctggtcagtctgggccgg
E N G Y D V Y H S P Q Y H F L V S L G R

      430     440     450     460     470     480
gcgaagagagccttcctgccaggcatgaacccacccccgtactccagttcctgtcccg
A K R A F L P G M N P P P Y S Q F L S R

      490     500     510     520     530     540
aggaatggagatccccctaatcacttcaacacccccataccacggcggcacacccggagc
R N E I P L I H F N T P I P R R H T R S

      550     560     570     580     590     600
gccgaggacgactcggagcgggacccccctgaacgtgctgaagccccgggccccggatgacc
A E D D S E R D P L N V L K P R A R M T

      610     620     630     640     650     660
ccggccccggcctctgttcacaggagctcccgagcgccgaggacaacagccccgatggcc
P A P A S G S Q E L P S A E D N S P H A

      670     680     690     700     710     720
agtgaaccattaggggtggtcggggcggtcgagtgaacacgcacgtgggggaacggggc
S D P L G V Y R G G R V N T H A G G T G

      730     740     750     760
ccggagggctgcgcgcccttcgccagttcatctag
P E G C R P F A K F I

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Mouse FGF-23 MLGTCLRLLVGVLC TVCS LGTARAYPDTSP LLGSMWGS LTHLYTATARTSYHLQIHRDGH 60
*** . *** * . ** . *** **** ** **** **
Human FGF-23 MLGARLRLWVCALCSVCSMSVLRAYPNASPLLGS SWGGLIHLTYTATARN SYHLQIHKNGH 60

VDGTPHQTIYSALMITSEDAGSVVITGAMTRRFLCMDLHGNI FGSLHFS PENCKFRQWTL 120
*** **** * **** * * **
VDGAPHQTIYSALMIRSEDAGFVVITGVMSRRYLCMDFRGNI FGSHYFDPENCRFQHQL 120

ENGYDVYLSQKHYYLVSLGRAKRIFQPGTNPPPF SQFLARRNEVPLLHFYTVRPRRHTRS 180
***** * * * * * *
ENGYDVYHSPQYHFLVSLGRAKRAFLPGMNPPPY SQFLSRRNEIPLIHFNTPIPRRRHTRS 180

AEDPPERDPLNVLKPRPRATPVVSCSRELPSAEEGGPAASDPLGVLRGRGDARGGAGG 240
*** * * * * * *
AEDDSERDPLNVLKPRARMT PAPASCSQELPSAEDNSPMASDPLGVVRGGRVNT HAGGTG 240

ADRCRPFPRFV 251
.. **** ..
PEGCRPFAKFI 251

FIGURE 4

00049967000

Figure 7

Codon usage for yeast (highly expressed) genes

AmAcid	Codon	Number	/1000	Fraction
Gly	GGG	33.00	0.86	0.01
Gly	GGA	70.00	1.82	0.02
Gly	GGT	2672.00	69.62	0.91
Gly	GGC	171.00	4.46	0.06
Glu	GAG	277.00	7.22	0.10
Glu	GAA	2442.00	63.63	0.90
Asp	GAT	1100.00	28.66	0.48
Asp	GAC	1211.00	31.55	0.52
Val	GTG	117.00	3.05	0.04
Val	GTA	75.00	1.95	0.03
Val	GTT	1548.00	40.33	0.56
Val	GTC	1026.00	26.73	0.37
Ala	GCG	36.00	0.94	0.01
Ala	GCA	203.00	5.29	0.06
Ala	GCT	2221.00	57.87	0.65
Ala	GCC	969.00	25.25	0.28
Arg	AGG	20.00	0.52	0.01
Arg	AGA	1336.00	34.81	0.83
Ser	AGT	116.00	3.02	0.05
Ser	AGC	94.00	2.45	0.04
Lys	AAG	2365.00	61.62	0.78
Lys	AAA	651.00	16.96	0.22
Asn	AAT	347.00	9.04	0.22
Asn	AAC	1259.00	32.80	0.78
Met	ATG	766.00	19.96	1.00
Ile	ATA	43.00	1.12	0.02
Ile	ATT	1223.00	31.87	0.52
Ile	ATC	1070.00	27.88	0.46
Thr	ACG	28.00	0.73	0.01
Thr	ACA	126.00	3.28	0.06

TABLE 7-36-1

Figure 7 (continued)

Thr	ACT	1129.00	29.42	0.50
Thr	ACC	962.00	25.07	0.43
Trp	TGG	325.00	8.47	1.00
End	TGA	10.00	0.26	0.09
Cys	TGT	254.00	6.62	0.89
Cys	TGC	33.00	0.86	0.11
End	TAG	11.00	0.29	0.10
End	TAA	85.00	2.21	0.80
Tyr	TAT	219.00	5.71	0.19
Tyr	TAC	913.00	23.79	0.81
Leu	TTG	2202.00	57.38	0.69
Leu	TTA	576.00	15.01	0.18
Phe	TTT	432.00	11.26	0.27
Phe	TTC	1145.00	29.83	0.73
Ser	TCG	26.00	0.68	0.01
Ser	TCA	149.00	3.88	0.06
Ser	TCT	1279.00	33.33	0.52
Ser	TCC	818.00	21.31	0.33
Arg	CGG	0.00	0.00	0.00
Arg	CGA	1.00	0.03	0.00
Arg	CGT	249.00	6.49	0.15
Arg	CGC	5.00	0.13	0.00
Gln	CAG	62.00	1.62	0.05
Gln	CAA	1225.00	31.92	0.95
His	CAT	236.00	6.15	0.35
His	CAC	433.00	11.28	0.65
Leu	CTG	52.00	1.35	0.02
Leu	CTA	236.00	6.15	0.07
Leu	CTT	90.00	2.35	0.03
Leu	CTC	14.00	0.36	0.00
Pro	CCG	10.00	0.26	0.01
Pro	CCA	1271.00	33.12	0.80
Pro	CCT	279.00	7.27	0.18
Pro	CCC	33.00	0.86	0.02

TABLE 9-95 (continued)

Circumstance	Percentage of Respondents (%)
Self-defense	95
To protect others	85
To protect property	75
To protect the community	65
To protect the environment	55

Amino Acid	Codon	Number	/1000	Fraction
Gly	GGG	6.00	0.28	0.00
Gly	GGA	380.00	18.04	0.22
Gly	GGI	575.00	27.29	0.34
Gly	GGC	746.00	35.41	0.44
Glu	GAG	1217.00	57.77	0.91
Glu	GAA	115.00	5.46	0.09
Asp	GAT	503.00	23.88	0.43
Asp	GAC	654.00	31.04	0.57
Val	GTG	719.00	34.13	0.45
Val	GTA	29.00	1.38	0.02
Val	GTT	226.00	10.73	0.14
Val	GTC	608.00	28.86	0.38
Ala	GCG	94.00	4.46	0.05
Ala	GCA	80.00	3.80	0.04
Ala	GCT	446.00	21.17	0.24
Ala	GCC	1277.00	60.61	0.67
Arg	AGG	48.00	2.28	0.06
Arg	AGA	12.00	0.57	0.01
Ser	AGT	16.00	0.76	0.01
Ser	AGC	267.00	12.67	0.23
Lys	AAG	1360.00	64.55	0.93
Lys	AAA	108.00	5.13	0.07
Asn	AAT	127.00	6.03	0.13
Asn	AAC	878.00	41.67	0.87
Met	ATG	387.00	18.37	1.00
Ile	ATA	4.00	0.19	0.00
Ile	ATT	390.00	18.51	0.29
Ile	ATC	969.00	45.99	0.71
Thr	ACG	114.00	5.41	0.08
Thr	ACA	34.00	1.61	0.02

Figure 8 (continued)

Thr	ACT	164.00	7.78	0.11
Thr	ACC	1127.00	53.49	0.78
Trp	TGG	243.00	11.53	1.00
End	TGA	1.00	0.05	0.01
Cys	TGT	20.00	0.95	0.08
Cys	TGC	220.00	10.44	0.92
End	TAG	12.00	0.57	0.17
End	TAA	58.00	2.75	0.82
Tyr	TAT	113.00	5.36	0.16
Tyr	TAC	574.00	27.25	0.84
Leu	TTG	210.00	9.97	0.12
Leu	TTA	9.00	0.43	0.01
Phe	TTT	62.00	2.94	0.09
Phe	TTC	635.00	30.14	0.91
Ser	TCG	195.00	9.26	0.17
Ser	TCA	29.00	1.38	0.02
Ser	TCT	103.00	4.89	0.09
Ser	TCC	558.00	26.49	0.48
Arg	CGG	7.00	0.33	0.01
Arg	CGA	25.00	1.19	0.03
Arg	CGT	281.00	13.34	0.34
Arg	CGC	465.00	22.07	0.55
Gln	CAG	703.00	33.37	0.91
Gln	CAA	66.00	3.13	0.09
His	CAT	88.00	4.18	0.22
His	CAC	312.00	14.81	0.78
Leu	CTG	1182.00	56.10	0.69
Leu	CTA	21.00	1.00	0.01
Leu	CTT	55.00	2.61	0.03
Leu	CTC	224.00	10.63	0.13
Pro	CCG	84.00	3.99	0.09
Pro	CCA	135.00	6.41	0.15
Pro	CCT	72.00	3.42	0.08
Pro	CCC	626.00	29.71	0.68

102020-89670860

Figure 9

Codon usage for enteric bacterial (highly expressed) genes 7/19/83

AmAcid	Codon	Number	/1000	Fraction
Gly	GGG	13.00	1.89	0.02
Gly	GGA	3.00	0.44	0.00
Gly	GGU	365.00	52.99	0.59
Gly	GGC	238.00	34.55	0.38
Glu	GAG	108.00	15.68	0.22
Glu	GAA	394.00	57.20	0.78
Asp	GAU	149.00	21.63	0.33
Asp	GAC	298.00	43.26	0.67
Val	GUG	93.00	13.50	0.16
Val	GUA	146.00	21.20	0.26
Val	GUU	289.00	41.96	0.51
Val	GUC	38.00	5.52	0.07
Ala	GCG	161.00	23.37	0.26
Ala	GCA	173.00	25.12	0.28
Ala	GCU	212.00	30.78	0.35
Ala	GCC	62.00	9.00	0.10
Arg	AGG	1.00	0.15	0.00
Arg	AGA	0.00	0.00	0.00
Ser	AGU	9.00	1.31	0.03
Ser	AGC	71.00	10.31	0.20
Lys	AAG	111.00	16.11	0.26
Lys	AAA	320.00	46.46	0.74
Asn	AAU	19.00	2.76	0.06
Asn	AAC	274.00	39.78	0.94
Met	AUG	170.00	24.68	1.00
Ile	AUA	1.00	0.15	0.00
Ile	AUU	70.00	10.16	0.17
Ile	AUC	345.00	50.09	0.83
Thr	ACG	25.00	3.63	0.07
Thr	ACA	14.00	2.03	0.04
Thr	ACU	130.00	18.87	0.35
Thr	ACC	206.00	29.91	0.55

TABLE 9-36-1983

[illegible]

Figure 10

Chromosomal localization of genes of the FGF family in human

Gene	Localization	Gene	Localization
FGF-1	5q31.3-q33.2	FGF-12	5q29-qter
FGF-2	4q26	FGF-13	X
FGF-3	11q13	FGF-14	13
FGF-4	11q13.3	(FGF-15)	
FGF-5	4q21	FGF-16	-
FGF-6	12p13	FGF-17	8p21
FGF-7	15q13-q22	FGF-18	5
FGF-8	10q25-q26	FGF-19	11q13.1
FGF-9	13q11-q12	FGF-20	3p21.3-p22
FGF-10	5p12-p13	FGF-21	12q13.1-qter
FGF-11	17	FGF-22	19p13.3
		FGF-23 □	12p13

Human FGF-15 gene has not been identified. The localization of human FGF-16 gene has not been determined.

Figure

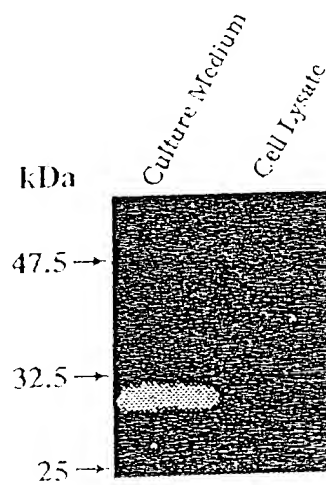


Figure 12

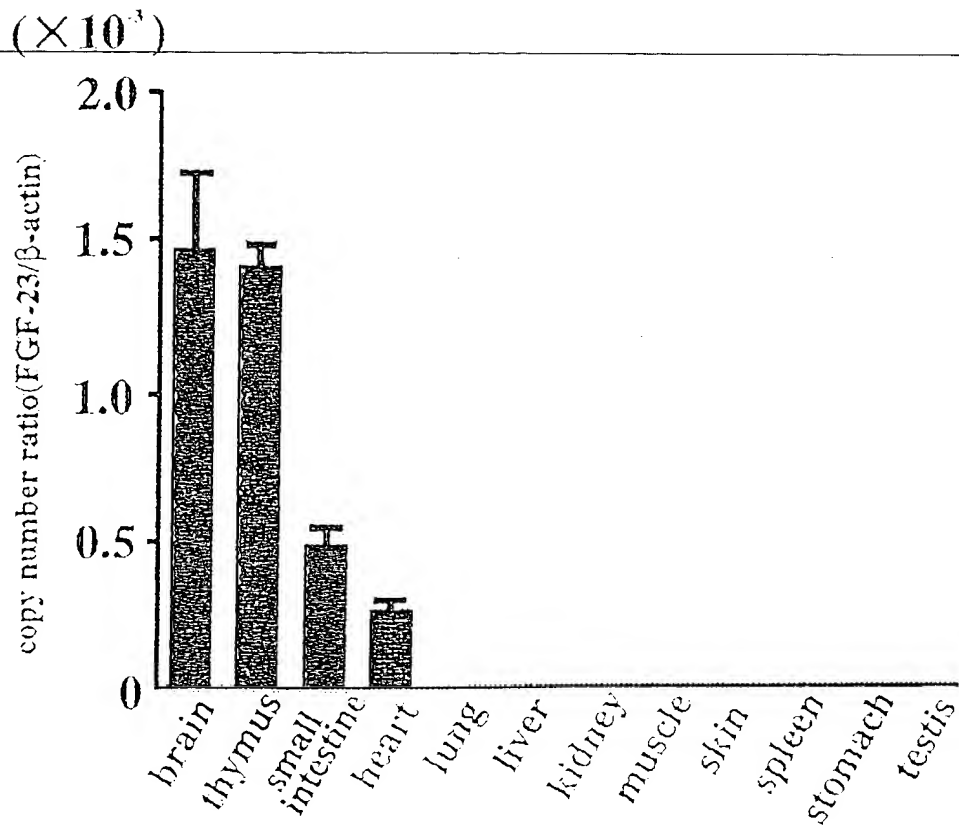
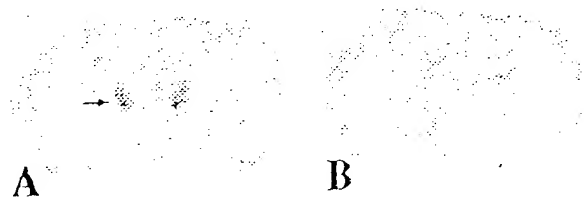
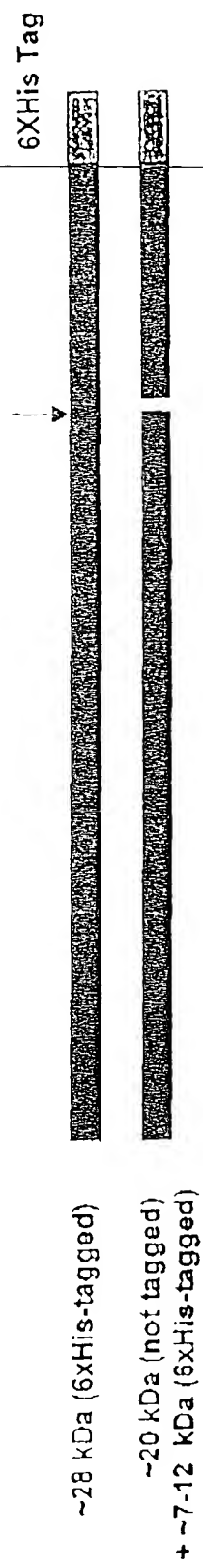


Figure 13



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T0000-896T0000

FIGURE 14



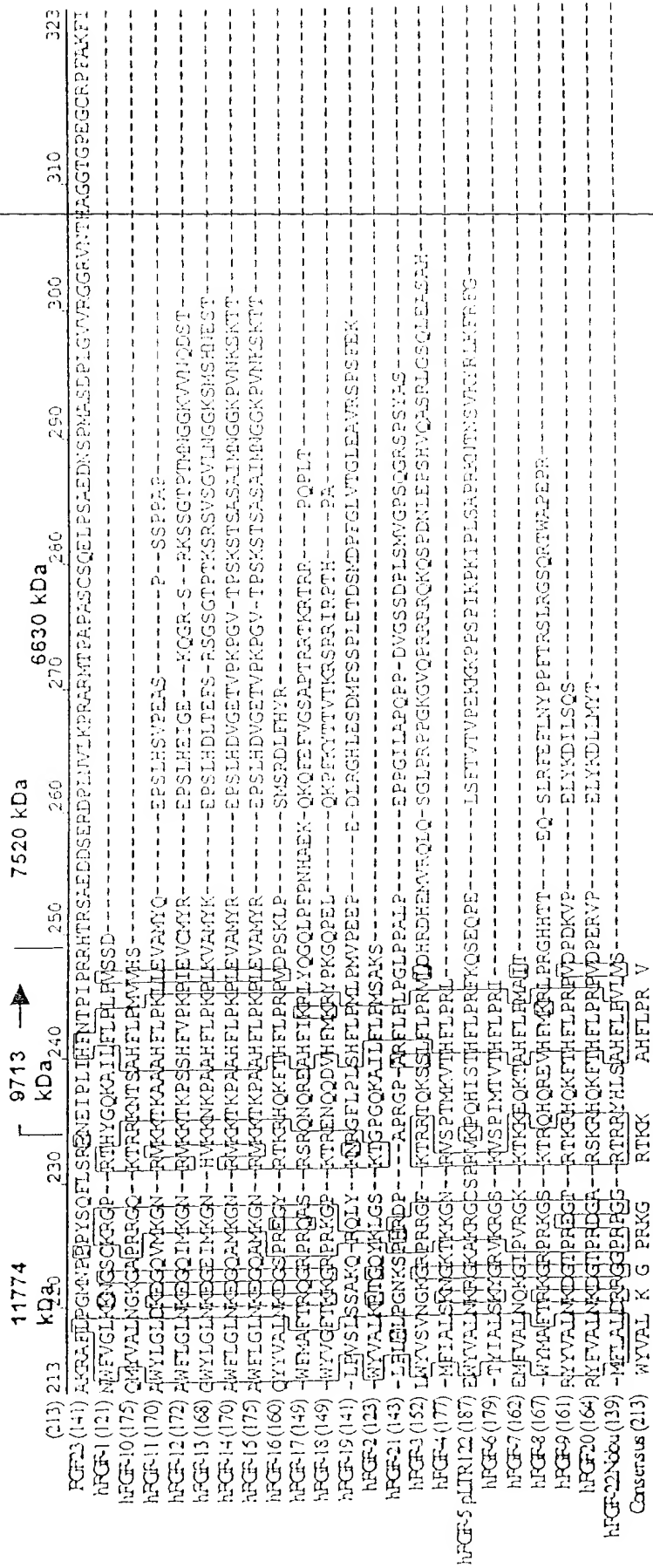


Fig. 15

Figure 16

kDa
250
98
64
50
36
30
16
6

Standards
hFGF-23
Standards

Cleavage of baculovirus-expressed
6XHis-tagged hFGF-23 secreted by Sf9 cells

N-terminal Sequence	Mass by Mass Spec	Sequence assignment	Cleavage Event(s)
NASPLLGS	17414	N27 – T178	<ul style="list-style-type: none"> Signal peptide removal (P26 ↓ N27) C-terminal cleavage (R179 ↓ S180) R179 removed by contaminating carboxypeptidase
XXWGGLIHLY	16761	S34-T178	<ul style="list-style-type: none"> Alternate Signal Peptide (G33 ↓ S34) C-terminal cleavage (R179 ↓ S180) R179 removed by contaminating carboxypeptidase
SAEDDSERDP	8204	S180-H256 (with SS bond)	<ul style="list-style-type: none"> C-terminal cleavage (R179 ↓ S180) H257 removed by contaminating carboxypeptidase Glycosylation present

Coomassie-Stained Gel

** aprotinin (added to preparation)